NUCLEAR PHENOMENA OF SEXUAL REPRODUCTION IN ANGIOSPERMS[1]

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WHAT constitutes sexual reproduction, fertilization or fecundation is so variously set forth in botanical literature that one naturally approaches a discussion of this subject with some timidity. Having been assigned the group angiosperms, in which a definition of sexual phenomena may be made more specific by concrete illustration, it seemed at the outset that a part of my task, at least, was simpler than that of some of my colleagues who take part in this section of our program; but a moment's thought convinced me that what might be gained from this limitation of my field was probably much less than the opportunities offered by the scope and diversity of phenomena in groups of lower plants.

In the preparation of this paper, the writer has kept clearly in mind the fact that the phenomena of sexual reproduction implies explicitly that a special significance is attached to the nucleus as in a large measure distinct from any function, or rôle of the cytoplasm; consequently he will deal first chiefly with nuclear behavior, leaving a discussion of the relation of nucleus and cytoplasm to be dealt with in a later paragraph.

Sexual reproduction in phanerogams implies the union of especially developed cells known as gametes, and the development of an individual plant from such union. While in this process the union of the nuclei is held to be more important, it is not inferred that the part taken by the cytoplasm is unimportant, but the writer does insist that the cytoplasm plays a secondary rôle in the

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most important results of sexual reproduction, namely, the transmission of parental characters. It will be maintained also that the union of mere gametophytic cells does not constitute fecundation, or a sexual process in phanerogams, nor is parthenogenesis—if such really exists in seed plants—to be confused with apogamy, nor apogamy with the various sorts of vegetative propagation of the sporophyte. Lastly, it is likewise the duty of the student of fecundation to consider the phenomena of graft hybrids which have been reported in recent literature, for such phenomena may have a profound significance in the shaping of future theories of sex and heredity, and on sex determination and control.

The problem of sexual reproduction in higher plants, and in lower ones as well, is complicated by the existence of two distinct phases, or individuals, in a life cycle, namely, the gametophyte and sporophyte; for it is necessary, in dealing with the rôle of the nucleus in sex, to consider a complete life cycle. Now gametophyte and sporophyte in phanerogams are fundamentally different hereditarily, and this difference is due chiefly to the fact that the former possess the haploid, or x number, of chromosomes, and the latter the diploid, or $2x$ number. Whatever respect or disrespect one may have for the "sacred" x and $2x$ number of chromosomes, the fact of their importance in any theory of sex and heredity remains the same, and must be taken into consideration. It is, of course, quite familiar to all that the change from sporophyte to gametophyte occurs in the formation of the spores, and that this change consists in the reduction of the number of chromosomes from the diploid, $2x$, to the haploid, x number; that this reduction is accomplished during the first mitosis in the spore mother-cells, this division of the nucleus being acknowledged as qualitative or differential. A detailed description of the evolution and behavior of the chromosomes during the tetrad divisions, and a discussion of controversies and of the different views held in regard to this evolution, or in regard to the differential character of any chromo-

some or set of chromosomes—are features which can not find consideration within the limits of this paper. I wish to state also that in the use of the expression, sexual reproduction, I shall keep in mind primarily, and for the sake of clearness, what will be called a complete sexual angiosperm, hereditarily speaking, that is, one with a complete life-cycle, embracing both sporophyte and gametophyte. Such an individual arises from the fusion of egg and sperm, each with the haploid number of chromosomes. The product of this fusion must develop into an adult sporophyte, capable of producing functional micro- and mega-spores, each with the haploid number of chromosomes. From this it will be seen that the mere fusion of gametophytic cells or nuclei is not necessarily regarded as a sexual process. It is to be understood also that the writer does not consider it imperative to stay within the confines of the foregoing definition, for like nearly all general definitions, this is made for convenience and for greater clearness in the general presentation.

No profounder statement has been made within the past half century than when it was said that the union of two sexual cells created a new individual—the individual with just twice the number of chromosomes as that possessed by either parent cell, namely, a sporophyte; for is it not the sexual process that makes possible all those phenomena understood by the expression, transmission of parental characters to offspring? And I mean especially the transmission of characters of two direct parents to each new generation. In this connection may we not ask also whether hereditary phenomena, such as engage the attention of biologists at the present day, exist among those simple plants in which sexual reproduction does not occur? Is there such a thing as phylogeny in plants that are without sex? The limits of this paper prohibit an attempt at an answer to the last two questions at this time. We shall have in mind then those characteristics which are handed down from parents to offspring. Some of these characters may mani-

fest themselves in the progeny with varying degrees of intensity, or in such combinations that entirely new marks or characters may appear; consequently characters are spoken of as dominant, recessive, latent, etc., and it not infrequently happens that estimates are placed upon absent characters. All of this implies that these external marks of living things are the manifestations of the activities of certain parts of the living substance, which are in competition, or among which there is an unceasing struggle. Attempts are also made to predict, by means of mathematical formulæ—and with surprising success in some cases—which of these forces or activities will gain the upper hand and dominate, and which will be secondary. Furthermore, sexual reproduction in phanerogams always implies a something that we know as maleness and femaleness, and this maleness and femaleness are phenomena which distinctively characterize the gametophytes. Maleness and femaleness may, in certain cases, give a distinctive mark to the sporophyte, as in diœcious plants, if we admit that such a thing as an absolutely diœcious sporophyte exists in angiosperms. But male and female marks are first manifested in the sporophyte of most seed plants by the production of the pollen and of the megaspores, which production is, of course, a matter of heredity. In the formation of pollen, it is well known that it is the nucleus which undergoes the important and complicated changes in division; the cytoplasm, so far as present knowledge extends, is halved arbitrarily. The same is likewise true in the development of the megaspores. In the case of each spore mother-cell, a qualitative division takes place, by which, as experimental evidence seems to indicate, different intensities of maleness and femaleness pass respectively to the several resulting cells. Both male and female gametophytes with their developing gametes grow under similar environmental conditions, namely, that of a parasitic habit within living tissue. The sperms of the pollen tube consist mainly of nuclei, with scarcely any distinctive characteristics in the scanty

cytoplasm. These nuclei contain the haploid number of chromosomes. In the development of the egg, the same number of chromosomes is strictly preserved, no matter what mitotic peculiarities may be observed in any other cells of the embryo-sac, apart from the egg-apparatus. The egg differs from the sperm in appearance chiefly in the amount of cytoplasm present, but the cytoplasm of the one is similar to that of the other. Apart from difference in shape, which is of no importance in phanerogams, the sexual nuclei reveal identical chromatin structures at the time of union, for both are in the resting, or non-mitotic state. Chromatin granules of the sperm nucleus mingle with those of the egg. When the nuclear membranes between the two contiguous nuclei have disappeared, it is not possible to distinguish paternal from maternal chromatin. It is not seen that chromatin particles of the sperm fuse or become paired with those of the egg nucleus. The gamete nuclei do not remain in any manner separate or distinguishable from each other, as in certain lower plants and in some animals, and one of the very interesting and important problems from the standpoint of hereditary considerations is the relation of male and female chromatin, during the life of the sporophyte. A number of authors have offered explanations of this relation based on observations made upon the first, or heterotypic, mitosis in the spore mother-cells, *i. e.*, at the end of the sporophytic cycle. Strasburger ('05) and Gregoire ('05), together with a number of their more recent students, maintain that the maternal and paternal homologous chromatin parts become associated in pairs previous to and during the synaptic balling up of the nuclear contents in the first mitosis of the spore mother-cells. This process, they assert, leads to the formation of two spirems (one paternal and one maternal) which become united side by side, to form the double chromatin thread, in which the homologous parental chromatin parts are brought near to each other.

The writer (Mottier, '07, '09) does not agree with this

view. From a careful and extended study of mitosis in spore mother-cells of higher plants, he is convinced that, confirming the observations of Farmer ('05) and others, there is no development of maternal and paternal spirems which unite laterally to form the double chromatin thread or spirem in the mitosis under consideration, but that the sporophytic chromosomes are arranged in a lineal series in the heterotypic spirem, and that consequently the two members of each bivalent chromosome are brought side by side, in case such an arrangement is attained by the two members of each bivalent, by a folding, looping or lateral approximation of parts of the spirem. In his own studies the writer is unable to find any justification of the doctrine that maternal and paternal chromatin is represented in definitely recognizable lumps designated by some observers as prochromosomes. While I can not agree with the view advocated by Strasburger and Gregoire, namely, the presynaptic or synaptic union of two spirems (male and female), because of my personal studies, I am also unable to accept their explanation upon the ground of theoretical considerations. Let us return for a moment to the fusion nucleus of the fecundated egg. It is perfectly clear that, soon after nuclear fusion, paternal chromatin elements, let us say, pangens, are indistinguishable from maternal elements. These pangens, assuming always the individuality of the chromosomes and of the pangens, correspond in form, size and staining qualities. The nucleolus or nucleoli of the egg are also similar to those brought in by the sperm, in case nucleoli are demonstrable in the sperm nucleus. There is nothing to lead one to believe that the parental pangens do not mingle in the resting nucleus (*i. e.,* not in mitotic activity). If there is a pairing of homologous parts, or exchange, inter-relation, or "Wechselwirkung" of pangens, or of any hereditary bearers, what reason is there to believe that such should not take place soon after fecundation, rather than at the close of the sporophytic ontogeny, or the beginning of gametophytic develop-

ment? If the nature and development of the sporophyte, from the standpoint of its inherited characteristics, is determined by what is transmitted to it by its parents, how may these parental tendencies operate unless they are intimately associated—unless some mutual relation, or a "Wechselwirkung" of the entities representing these tendencies, is in continuous activity? Furthermore, when the fusion nucleus of the fertilized egg in angiosperms divides, the spirem separates by cross segmentation into the $2x$ number of chromosomes, x being male and x female. These sporophytic chromosomes are arranged in lineal series, or end to end, to make the spirem, which splits longitudinally. I do not believe many cytologists will contend that the sporophytic spirem is formed by the lateral coming together of male and female spirems. If the parental chromosomes are arranged tandem to form the sporophytic spirem, why should they be arranged in any other manner to form the heterotypic spirem?

Returning now to the fecundated egg, it is seen that the fusion nucleus presents the same visible structure as that of either gamete, with the exception that an additional nucleolus or nucleoli may sometimes be observed. The essential demonstrable act in this fusion concerns the nuclei; the behavior of the cytoplasm that may accompany the sperm nucleus is largely a matter of conjecture, for it is not possible to trace its behavior with any degree of accuracy, either in the living state, or by means of the indirect method of study. However, to satisfy the demands of the most radical we may admit that sperm cytoplasm unites with egg cytoplasm in the act of fecundation. I have described in some detail the structural union of the sexual nuclei; for in a later paragraph will be discussed the relative significance of nucleus and cytoplasm in sexual reproduction, and as factors in the transmission of hereditary characters.

One male nucleus of the pollen-tube is concerned in sexual reproduction as that term is understood in this paper, but as the union of the second male nucleus with

the polar nuclei in a number of plants has been associated with, if not actually regarded as, a sexual act, the phenomenon will receive a brief mention. In 1897, the writer (Mottier, '97) called attention for the first time to the fact that the second male nucleus applied itself to one of the polar nuclei in *Lilium Martagon,* and shortly after that date the actual union of this male nucleus with the endosperm nucleus of the embryosac was reported for various plants by different observers. Guignard spoke of this nuclear union as a second fecundation, hence arose the idea of "double fecundation" in phanerogams. The fusion of the two polar nuclei and the second male nucleus resembles physically the real sexual union, as do also nuclear fusions in ordinary vegetative cells wherever such may occur. It recalls a sexual act in subsequent behavior, for it is maintained that endosperm thus arising is of a hybrid character, and that this hybrid character is due to the hereditary influence of the male nucleus. The hereditary influence of the second male nucleus upon the endosperm we may admit for the sake of argument, for whatever else this phenomenon may signify, it certainly shows the greater importance of the nucleus in the transmission of characters. But, that this union of nuclei in the endosperm cell is not a sexual process, as defined in a foregoing paragraph, is seen in the fact that the endosperm is merely a continuation of the female gametophyte, developed subsequent to fertilization of the egg, and for the nourishment of the sporophyte, just as a fern prothallium may continue its development, following the fecundation of an egg and its subsequent development into the embryo fern.

In a group of organisms in which a structure or function is so universally present, the absence of the same in any one or several of such organisms elicits at once our attention, and, in this respect, apogamy and parthenogenesis become of special interest. Although apogamy and parthenogenesis do not involve the sexual act, yet an accurate and intimate knowledge of these

phenomena is likely to modify profoundly our views of sexual reproduction, especially if a series of generations of apogamously produced plants be compared with a series of sexually produced individuals, of the same or related species, in regard to the subject of variation, individual vigor, manner of propagation, transmission of certain characters, etc. The term apogamy is here used to signify the development into an embryo of the egg-cell possessing the double or $2x$ number of chromosomes without the union with a sperm nucleus from the pollen-tube (somatic parthenogenesis of Winkler, parthenopogamy of Farmer and Digby). It is not deemed desirable to connect the word parthenogenesis with such a process, for a reproductive cell, although developed morphologically as a gamete, is not so considered unless it contain the reduced number of chromosomes. Accordingly, the term parthenogenesis will be applied only to the development into an embryo sporophyte of an egg containing the x number of chromosomes.

In recent years several notable cases of apogamy among phanerogams have been described, among which may be mentioned *Antennaria alpina* by Juel (1900), species of *Alchimilla,* especially of the group Eualchimilla by Murbeck and Strasburger ('04), *Taraxacum officinale* by Juel, *Wikstroemia,* by Winkler, together with several others from different families of plants. In *Antennaria alpina,* for example, the tetrad divisions do not take place in the megaspore mother-cell which functions at once as the megaspore. Naturally this cell contains the diploid number of chromosomes. From this cell there develops an apparently normal embryo sac, with the exception that the polar nuclei do not unite. The cell which represents the egg develops without fecundation into an embryo sporophyte. The process in the other species mentioned is in the main similar to that of *Antennaria alpina,* differing only in certain details, which may not be enumerated here. While, in these apogomous species, an apparently normal gametophyte develops, it may be very seriously questioned

whether such embryosacs, whose egg-cells contain the these apogamous species, an apparently normal gametophytic. However, progress in science is not accomplished by controversies and discussions of terminology. The main thing which interests us here is the effect, from an hereditary standpoint, that apogamy, of the sort mentioned, has upon the species so affected. True it is that such apogamous progeny have the characters of both parents, male and female, but it is a remote parentage. There is no new parent introduced with each new generation, that is, each time a plant comes from a seed, and it seems not improbable that the loss of certain very important reproductive functions may be expected in later generations of such plants. In reality certain observed facts seem to bear out this suggestion, as Strasburger found that, in some of the Eualchimillas, degeneration took place in the pollen mother-cells before the spores were fully formed. Whether apogamously developed plants will behave in a manner similar to those propagated vegetatively, *e. g.*, by cuttings, or as normal sexually produced individuals, future research must determine. Until the observations of the several observers have been confirmed by others, and until experimental cultural studies are made to ascertain the behavior of apogamous plants along with those possessing sexuality, speculation seems idle.

In regard to parthenogenesis as defined in a preceding paragraph, this phenomenon is claimed to occur in certain species among rather widely separated families. The best known instances are *Thalictrum Fendleri,* as reported by Day (1896), *Thalictrum purpurescens,* by Overton (1902, 1904) and *Wikstrœmia indica,* by Winkler (1904, 1905). However, for *Thalictrum purpurescens,* Overton (1904, p. 278) expressly states that in some cases no reduction in the number of chromosomes occurs in the embryosac mother-cells, and that tetrads are not formed, so that apogamy certainly occurs in this species also. In regard to *Wikstrœmia* it may be added that the recent investigations of Strasburger upon this

plant and other Thymelaceæ, point rather towards apogamy than parthenogenesis.

Even more perplexing to the student of sex and heredity than apogamy or parthenogenesis are the phenomena presented by what are known as graft hybrids. A number of seed plants of a hybrid nature are known to botanical science, which have not arisen by means of seed production, but presumably from the callus formed at the juncture of the stock and scion in grafting. The most noted of these is, of course, *Cytisus Adami,* which is supposed to have arisen from *Cytisus laburnum* and *Cytisus purpureus* as a graft hybrid. This problem, which has held, in a large measure, the interest of biologists for about eighty years, seems now to be on a fair way towards a solution, having as a starting point the production of a graft hybrid experimentally. Hans Winkler, as is well known, has produced a plant which, in point of flower, fruit and foliage, seems to be a hybrid between the common nightshade *Solanum nigrum* L. and the tomato, *Solanum lycopersicum* L. of the King Humbert yellow-fruited variety, by an ingenious method of grafting in which the nightshade was used as the stock and the tomato as the scion. Perhaps a very brief statement of the process may not be out of place here.

Using the cleft method of union, Winkler grafted vigorous shoots of the seedling tomato upon the stem of the nightshade. As soon as union had taken place the scion was cut off near its base in such a way that the apical cut surface consisted partly of nightshade and of tomato tissue. Of the adventive shoots arising only those which sprang from along the line of union of the two specifically different tissues were allowed to grow. In one particular case fourteen of such sprouts were removed and transplanted as cuttings. Of these eight proved to be *Solanum nigrum,* five pure *Solanum lycopersicum,* and one the hybrid in question. This plant grew to flower and fruition, and as stated in the foregoing, revealed hybrid characters in stem, leaf, flower and fruit. This hybrid Winkler named *Solanum*

tubingense H. Wklr. (*S. nigrum* L. + *S. lycopersicum* L., 1908). For a detailed description of the plant the reader is referred to the original publication.[1] That the conditions under which such a graft hybrid is produced are very rarely fulfilled, is seen in the fact that from the 268 graftings made by Winkler in 1908, 3,000 adventitious shoots were developed after decapitating the graft in the manner described, and of these 3,000 the vast majority were specifically pure; five were chimeras, and one the hybrid referred to. Now the all-absorbing question for the cytologist is: By what means are parental characters transmitted in cases of this sort? Of course speculation is futile until the histological facts are known, but, assuming that such shoots are real hybrids, two guesses may be offered. There may have been (1) a migration of nuclei from cell to cell and their subsequent fusion, as in certain fern prothallia developing apogamous sporophytes, or (2) the hereditary transmission may have been accomplished by cytoplasmic union between cells, or by some sort of enzyme action.

It is highly probable that these remarkable adventitious shoots are not true hybrids, but mere chimeras.

In a personal communication, Dr. Winkler has very kindly informed me that seeds of "*Solanum tubingense*" produced pure nightshades (*Solanum nigrum*), and those of "*Solanum proteus*" pure tomatoes (*Solanum lycopersicum*).

The fact that seeds of "*Solanum tubingense*" produced pure nightshades seems to be conclusive evidence that the structure in question is not a hybrid, but merely a remarkable chimera. Strasburger ('09) has just published the results of a histological study of the tissues formed at the juncture of stock and scion in grafts of *Solanum nigrum* and *Solanum lycopersicum,* and he reports that neither nuclear migrations from one vegetative cell to the other nor nuclear fusions in any of these cells were observed. These results are in accord with the same author's cytological studies on other supposed

[1] *Ber. d. Deutsch. Bot. Gesellsch.,* 26*a*: 595–608, 1908.

graft-hybrids. Winkler's histological studies on "*Solanum tubingense*" have not yet been made public, but all the facts thus far seem to indicate that so-called graft-hybrids, including *Laburnum Adami,* the *Bizzarrias* and those of *Mespilus,* etc., are only vegetable chimeras.

At the juncture of stock and scion in grafts, especially in the case of those that produce adventitious shoots of such remarkable character, there is a cell-complex formed of the vegetative cells of two specific individuals, and the specifically different cells may be regarded as being so intermingled and reacting upon each other in such a manner as to produce adventitious shoots of an almost exact hybrid character in so far as vegetative marks are concerned. In the case of Winkler's "*Solanum tubingense,*" whose seeds gave only pure nightshades, it is clear that both egg and pollen were descended from pure nightshade cells, as the nightshade and the tomato do not cross.

Although the problem of the so-called graft hybrids can not be regarded as definitely settled, yet nearly all the facts go to strengthen the view that hybrids are formed only by the union of cells and nuclei sexually differentiated, and that fecundation and the transmission of characters are not accomplished by the protoplasm in general, nor by the action of an enzyme, nor is it the expression of metabolism, but by the *union of specific material entities in the sexual nuclei.*

Although the concensus of opinion among biologists attributes to the nucleus by far the most important rôle in the process of sexual reproduction in its fullest significance, yet there is still some difference of opinion in regard to the relative functions of nucleus and cytoplasm in imparting the stimulus to growth and cell division, and in the transmission of parental characters—the two chief constellations of phenomena following the sexual act.

To arrive at any satisfactory conclusion in the light of existing literature, a careful analysis of cell structure and of the functions more directly concerned is neces-

sary, both from the standpoint of phylogeny and from that of the individual, or individuals concerned. Careful investigations of recent years upon the cells of certain lower plants seem to justify the opinion that the more original or primary protoplasm is to be conceived as being entirely without organized nuclei, possessing uniformly in all parts its formative and nutritive functions. Then there gradually came about, phylogenetically speaking, a separation of the constructive, nutritive—and may we also say—directive functions in this substratum, those parts of the plasm having formative activities being the first differentiated bearers of hereditary characteristics. These particles or granules may have remained for a long time distributed in the general plasmic mass, just as we find in certain existing Cyanophyceæ and bacteria "chromatin bodies" distributed throughout the cell rather than collected in a typical nucleus. The next step in the evolution of more differentiated protoplasm occurred when the formative parts, be they known as chromatin bodies, or what not, became separated from the surrounding plasm by a membrane, or, in other words, the creation of nucleus as distinct from cytoplasm. In the light of known facts no one, I think, will seriously believe that among the lower plants the nucleus is as highly differentiated as among seed plants, consequently a larger number of functions must have been performed by the various hereditary units, and a much simpler method of nuclear division demanded. This view seems well borne out by the simpler method of nuclear division in certain lower plants and in cells of higher plants, which have taken on a purely vegetative rôle, and which divide by the direct method or fragmentation. As soon, however, as differentiation in the hereditary units increased, a much greater complexity in the mechanism of division followed, a conclusion to which the mitotic phenomena in higher plants stand as incontestible testimony. On the other hand, we do not mean to imply that progressive differentiation was confined to the nucleus alone, for the cytoplasm of higher plants reveals

evidence of unmistakable differentiation. I do not allude to the alleged hereditary substance, chondrosomes, to be mentioned beyond, but merely to such differentiation as spindle fibers, which in many higher plants are almost wholly of cytoplasmic origin. As is well known, Strasburger has endeavored to make things clearer by applying to such parts of the cytoplasm as spindle fibers centrosomes, centrospheres and the plasma membrane of the cell, the term kinoplasm, attributing to this substance certain activities. The researches of Noll upon marine algæ indicate with a very high degree of probability that the plasma membrane is the part of the protoplasm which takes a leading part in responding to external stimuli. The doctrine that an enucleated cell can not do any constructive work, as, for example, forming a cellulose wall, has become so generally accepted that the same has found its way into general reference works. This doctrine has in recent years been disputed, but so far as I am aware it has not been satisfactorily disproved. I shall not bring into this category such cytoplasmic differentiations as chloroplasts and other plastids, but enough has been said to indicate that the cytoplasm as well as the nucleus is a differentiated body, which means a diversity of functions or activities. Now, although cytoplasm and nucleus have certain functions that seem in a large measure independent, yet the interrelation of these two parts of the cell is such that neither can exist and function to any great extent without the other. No one has up to the present time been able to isolate a nucleus and keep it alive any length of time apart from living cytoplasm. Whatever the nucleus does, it must do in connection with living cytoplasm. The cytoplasm is in a sense the special environment of the nucleus, and it is in this environment that the nucleus must exist and function. It is also reasonable to believe that, in certain functions of the nucleus, the cytoplasm acts largely as an environmental factor. It is, however, an environment so intimately connected with the nucleus that even a momentary separation may prove fatal, for the skill of

the experimenter in this field is yet to be demonstrated.

On the other hand, while the cytoplasm may exist for some time wholly apart from a nucleus, and although during this separate existence the cytoplasm may respond to certain stimuli, yet it can not do constructive work—a phenomenon which seems to indicate roughly the chief province of these two parts of the living cell.

In the light of the foregoing analysis, we may now consider some results obtained by indirect methods, and through certain experiments.

Since the declaration of O. Hertwig in 1875, that fecundation consisted essentially in the union of an egg and a sperm nucleus, this doctrine has received general acceptance. Some observers maintain that this idea places undue emphasis upon the importance of the nucleus, and claim that the cytoplasm is almost of equal significance. In a recent publication Meves ('08) describes in great detail rod or thread-like bodies in the cells of the very young embryo of the chick, which he designates as chondriosomes, and which he regards as cytoplasmic bearers of hereditary characters. The same author in 1904 described and figured similar rods and threads as occurring abundantly in the tapetal cells of *Nymphæa alba*. In the tapetal cells of the anther of *Ribes Gordonianum,* Tischler ('06, p. 573) calls attention to slender rods of varying length, which he designates as chromidial substance, stating that they came out of the nucleus. The writer has examined many thousands of tapetal cells from various plants, fixed and stained in a manner quite similar to that used by Meves, but no such bodies have been found as those figured by this author. It is not my intention to discuss this phase of the subject from the standpoint of zoological literature, but it may be said that tapetal tissue is not the place that a botanist would go to look for especially differentiated hereditary substance. If hereditary substance, such as Meves attributes to the cytoplasm of tapetal cells, really exists, it seems very strange to a plant cytologist that it can not be demonstrated in spore mother-cells, where,

above all parts of the plant, protoplasmic structures are most clearly brought out.

As stated in the foregoing, fecundation manifests two constellations of phenomena, the transmission of parental characters and the power of growth and division of the egg. That these two categories are in a measure distinct is amply attested by phenomena of common observation, and by experimental evidence. That various sorts of environmental stimuli, from the sting of an insect to the increased osmotic power of surrounding water, will impart to living cells the power of growth and division is well known. The sting of an insect, for example, will stimulate growth and cell division in stem and leaf, which results in a gall; the presence of a pollen tube will induce an ovule to grow to mature size though no embryo develops within it. In these and in other similar cases, too numerous to mention, we have merely responses to external stimuli, for doubtless the pollen tube may act as an external stimulus, and no one will contend that these phenomena have to do with the transmission of parental characters. From our standpoint the phenomenon of artificial parthenogenesis merits especial attention. When the egg-cells of certain marine animals are stimulated to develop by external agencies of whatever sort, it has become fashionable to speak of the fact as fertilization, but whatever meaning be put into the word fertilization, the phenomenon in question is not fecundation or sexual reproduction. Even though in every case the most sanguine expectations of the experimenter be realized, namely, the development into an adult of an egg thus stimulated, the process would teach us nothing more about sexual reproduction and the transmission of parental characters than ordinary parthenogenesis. The fact that a larva having purely maternal characters will develop from a sea-urchin egg with which the sperm of a starfish had united, does not show that hereditary characters are handed down by the cytoplasm. If, on the contrary, the gastrula, showing only maternal characters, which Godlewski ('06) reared

from the union of an enucleated egg-fragment of the sea-urchin and the sperm of a crinoid, could have developed into an adult, or even into the larval stage, which still revealed only maternal characters, the cytoplasm might have regained some of its old-time prestige, but even then it is doubtful whether that fact would have wrested from the nucleus its monopoly as a transmitter of parental characters.

In such cases as the embryo hybrid of sea-urchin and starfish, mentioned in the foregoing paragraph, it seems very probable indeed that the cytoplasm of the egg directs and even controls the growth of the embryo for a short time subsequent to fecundation, but it is very improbable that the cytoplasm does more. Even in the case of a complete fusion of the egg and sperm nuclei, this and similar experiments seem to indicate only the dominance of the egg nucleus over that of the strange sperm. With the egg nucleus operating in its own special environment, and attributing a directive or regulative function to the cytoplasm, the result is what might reasonably be expected. That the cytoplasm is differentiated, and that it directs or regulates the formation of certain parts of the embryo in some animals, is clearly shown by the various interesting and important studies of cell lineage carried out by Conklin ('05) and others. Important and far-reaching as are these studies in contributing to our knowledge of living protoplasm, they do not teach us very much concerning the cytoplasm as an heredity bearer, nor do I understand that the respective observers make such claims.

The stimulation of egg-cells to growth and division by immersing them in water having different physical or chemical properties than their normal surroundings, or by injecting chemicals into the ovaries or ovules by means of an hypodermic syringe, are lines of study that are valuable and interesting in showing the response of living cells and tissues to external stimuli, for it is the business of the physiologist to know what cells can do under any and all conditions; but that these experiments

have anything to do with sexual reproduction or in elucidating the more fundamental principles in the evolution of organisms connected with sexual reproduction, still remains to be seen.

On the other hand, if one regards the elementary life processes merely as the expression of metabolism, then hereditary peculiarities are only the expression of metabolism. That which is inherited is for each organism only that kind of metabolism peculiar to the organism. Of course, the writer can not subscribe to this view. Neither does he maintain that the cytoplasm takes no important part in sexual reproduction. He has called attention to the opinion that the stimulus to growth and cell division which follows every sexual act fully accomplished, and which may be brought about apart from the act of fecundation, has been confused with the main result of sexual reproduction, namely, the transmission of parental characters. It is held that the present state of our knowledge still maintains the doctrine that the "monopoly" of transmitting hereditary characters still belongs to the nucleus, and that these hereditary parental characters are represented in the nucleus by material entities. It matters little whether we speak of these material representatives as pangens, or what not. The opinion is expressed that the chief function of the cytoplasm, apart from purely nutritive activities, in its relation to the nucleus is directive or regulative in the sense of being responsive to external stimuli. In so far as the transmission of parental characters go, the cytoplasm plays about the same rôle compared with the nucleus as the environment does in the development of the individual organism.

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